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Short-latency sensory afferent inhibition: conditioning stimulus intensity, recording site, and effects of 1 Hz repetitive TMS

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Maximilian Fischer
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Prüfungsausschuss, zweite/r Gutachter/in: **Prof. Dr. Jens Fiehler**

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PAPER

Short-latency sensory afferent inhibition: conditioning stimulus intensity, recording site and effects of 1Hz repetitive TMS

M Fischer, M Orth MD PhD

Department of Neurology, Universitätsklinikum Ulm, Oberer Eselsberg 45/1, 89081 Ulm, Germany

For correspondence:
Dr M Orth
Department of Neurology
Universitätsklinikum Ulm
Oberer Eselsberg 45/1, 89081 Ulm, Germany
Tel.: 0049 731 50063095
Fax: 0049 731 50063082
Email: michael.orth@uni-ulm.de

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1.1 Abstract

Background: The transcranial magnetic stimulation (TMS) paradigm short-latency sensory afferent inhibition (SAI) investigates sensori-motor integration. Conventionally, one stimulation intensity is used for the conditioning pulse to the peripheral nerve,.

Objective/hypothesis: To examine the variability, the dimension of stimulus intensity and recording site in SAI.

Methods: In 17 healthy individuals three peripheral nerve stimulation intensities were used: Just above sensory threshold, just above motor threshold, and in between. Motor evoked potentials (MEPs) and long-loop reflexes were recorded from first dorsal interosseus (FDI) and abductor pollicis brevis (APB) before and after repetitive motor cortex TMS (1Hz, 1800 stimuli at 95% resting motor threshold).

Results: Between-subjects variability of SAI was higher than variability between sessions. Median, or ulnar, nerve stimulation decreased MEP size in FDI and APB at inter-stimulus intervals of N20, N20+2 and N20+4. Only with median nerve stimulation MEP size increased in APB, but not FDI, at N20+8 to N20+16. These effects increased with increasing stimulation intensity. RTMS reduced MEP size but had no effect on SAI, or transcortical reflexes.

Conclusions: Effects on MEP size in SAI depend on stimulus intensity and are not limited to anatomically homotopic muscles. Inhibitory rTMS modulates motor output but not the interaction of sensory inputs with the motor cortex.

1.2 Introduction

Sensori-motor integration describes the process by which the sensory and motor systems communicate and coordinate their activities. It involves the reception of a stimulus, e.g. tactile, acoustic, or optical, and its transmission to the central nervous system where the stimulus is interpreted. In case of a motor response this involves the transmission of impulses along corticospinal motoneurones to a group of muscles, which elicits an appropriate movement.

To understand the physiology of sensori-motor integration many studies have used transcranial magnetic stimulation (TMS) methods. These examine the effect of an afferent sensory input from the hand on the excitability of human motor cortex. Motor evoked potentials (MEPs) are modulated by a preceding electrical stimulus to mixed nerve [1-3] or cutaneous nerves [4-7]. Since the peripheral nerve stimuli have no effect (at the same latencies) on responses evoked by transcranial electrical stimulation or on F-waves [3, 8-10] they likely origin at a cortical level. There are suggestions that these effects have a somatotopical organisation since the largest changes in MEPs are seen in muscles nearest the site of stimulation [9, 10].

A paradigm frequently used is short-latency sensory afferent inhibition (SAI). Here an electrical stimulus given to a mixed nerve, most commonly the median nerve, above its motor threshold precedes the TMS pulse to the motor cortex. At inter-stimulus intervals slightly longer than the N20 component of somatosensory evoked potentials (SEPs) this reduces the size of the motor evoked potential (MEP) [3, 11]. In addition, at longer inter-stimulus intervals between conditioning stimuli and TMS shock to the motor cortex the MEP size increases [11]. In the present study, we extended the investigation of SAI in three ways: 1) we investigate coefficients of variation between sessions and between subjects; 2) we examined the effects of different intensities of mixed nerve stimulation on MEP size; 3) we stimulated two mixed nerves, median and ulnar, and recorded simultaneously from first dorsal interosseus (FDI) and abductor pollicis brevis (APB) muscles to assess the differential effects on anatomically

homotopic, or heterotopic, muscles. In a second experiment we stimulated the motor cortex using an inhibitory repetitive TMS protocol to examine the effects on MEP size, SAI, long-loop reflexes and the relay time from somatosensory cortex to motor cortex [12, 13]. A previous study had shown some subtle inhibitory effects of a similar inhibitory rTMS protocol on somatosensory cortex excitability assessed using somatosensory evoked potentials (SEP) [14], and the amplitude of long latency reflexes [15] while facilitatory rTMS increased the cortical component of the SEP and long latency reflexes [15]. Our hypothesis was that the inhibitory effects of rTMS conditioning extend beyond the motor cortex (MEP reduction) and the somatosensory cortex (SEP) to involve the integration of sensory stimuli with motor output. To test this we examined the effects of rTMS on SAI with the afferent conditioning stimulus given to the median, or the ulnar, nerve.

1.3 Material and Methods

1.3.1 Participants

Seventeen healthy right handed (Edinburgh handedness inventory, [16]) Caucasian volunteers were studied (mean age 24, range 23-27, 5 women). Subjects were asked to refrain from caffeine-containing beverages on the day of the experiment. All participants took part in experiment one; 16 participants took part in the second experiment. Participants gave informed written consent, and the local ethics committee approved the study protocol.

1.3.2 Electromyography recordings

Surface electromyograms (EMG) were recorded from the right first dorsal interossoeus (FDI) and the right abductor pollicis brevis (APB) muscle using silver/silver-chloride disc surface electrodes (1 cm diameter) in a belly tendon montage. The EMG signal was amplified and analogue filtered (30Hz to 1kHz) with a Digitimer D150 amplifier (Digitimer Ltd., Welwyn Garden City, UK). Data (sampling rate 4kHz) is digitised for off-line analysis using Signal software (Cambridge Electronic Devices, Cambridge, UK).

1.3.3 Transcranial Magnetic Stimulation

Participants were seated in a comfortable chair. They were asked to relax as much as possible. Magnetic stimuli were given with a hand-held figure-of-eight coil (outer winding diameter 9cm) connected to a High Power Magstim 200 stimulator (Magstim Co., Whitland, Dyfed, UK). This stimulator generates a magnetic pulse with monophasic waveform and induces a current in the brain with posterior-anterior flow when the coil handle is positioned at an angle of 45° pointing backwards. The optimal spot for right FDI and APB stimulation was marked with a felt pen.

RTMS (1Hz, 1800 stimuli to the left motor cortex at 95% resting motor threshold) was applied using a Magstim Rapid (Magstim Co., Whitland, Dyfed, UK) which generates a biphasic (posterior-anterior/anterior posterior) current flow in the brain. All the stimulation variables followed the published safety guidelines [17, 18].

1.3.4 Motor thresholds

Resting motor threshold (RMT) was defined as the minimum intensity needed to evoke an MEP of >50μV in 5 out of 10 consecutive trials in the relaxed FDI. Active motor threshold (AMT) was defined as the minimum intensity (in % of maximum stimulator output) needed to evoke a MEP of >200μV in 5 out of 10 trials in the tonically active FDI (~20% of maximal contraction as assessed visually on an oscilloscope). Thresholds were approached from above threshold in steps of 1% stimulator output. Once no MEP could be elicited the intensity was increased in steps of 1% stimulator output until a minimal MEP was observed. This intensity was taken as motor threshold.

1.3.5 Somatosensory evoked potentials

After stimulation of the median or ulnar nerve with surface electrodes at threshold intensities for motor stimulation somatosensory evoked potentials were recorded with a needle electrode over the somatosensory cortex (2cm posterior of C3 in the international EEG 10-20 system) referenced against the opposite ear lobe. Stimulation was given at a frequency of 3 Hz; a total of 300 stimuli were averaged.

1.3.6 Short latency afferent inhibition (SAI) by somatosensory input from the median, or ulnar, nerve

SAI of the motor cortex was examined as previously described [3]. In brief, a MEP of ~1mV peak-to-peak amplitude is elicited in the FDI by TMS. A paired pulse paradigm examines the influence on MEP size of a supra-threshold electrical stimulus given to the median nerve through bipolar surface electrodes with the cathode proximal to the anode. The electrical stimulus to the median nerve was delivered at three different intensities: above sensory threshold when participants had a sensation in the distribution of the median nerve, just above the threshold to elicit a visible contraction in the thenar muscles, and at an intensity between sensory and motor threshold. To the ulnar nerve, electrical stimulation was delivered above sensory threshold and above motor threshold. Since the difference between sensory

and motor threshold was smaller than for the median nerve we did not use a third intensity between sensory and motor threshold. Stimulation preceded the TMS pulse to the FDI hot spot in relation to the N20 component of sensory evoked potentials (N20, N20+2, N20+4, N20+6, N20+8, N20+10, N20+12, N20+14, N20+16). Twenty trials of the MEP elicited by TMS alone and 10 trials of conditioned MEPs for each inter-stimulus interval (ISI) were collected. The amplitude of the MEP in the FDI, or APB, was measured with in-house software. The intensity of the TMS pulse was adjusted to result in a MEP of between 0.5 and 1mV in amplitude. The intensity was adjusted again following rTMS so that unconditioned MEP amplitude was similar before and after rTMS. The average amplitude of the conditioned MEP was expressed in percent of the average amplitude of the un-conditioned MEP alone. Trials recorded while the patients contracted the hand muscles were excluded on-line. No trials were excluded in the off-line analysis.

1.3.7 Long-latency reflexes

The method followed the description by Deuschl & Eisen [12]. Reflexes were elicited in the contracted right abductor pollicis brevis muscle by electrical stimulation of the median nerve at the wrist. Subjects sat with their pronated forearm supported before them on a table and contracted the APB muscle isometrically to approximately 40 % of maximum by abducting the thumb against a force transducer with reference to a visual display before them. The median nerve was stimulated at motor threshold intensity using surface electrodes with the cathode proximal to the anode (stimulus duration, 1.0 ms; random rate from 0.9 to 1.1 Hz; constant current source). We visually inspected the reflexes following electrical nerve stimulation on average records of full-wave rectified EMG activity. Then we determined the end of the short latency and beginning of the long latency reflex when the average surface rectified EMG increased abruptly at a latency of between 45 and 55 ms. First, we estimated the duration of the short and long latency components of the EMG responses. Then, we calculated the integral of the rectified EMG activity as the size of the reflexes.

1.3.8 Data analysis

Peak to peak amplitude of MEP were measured with in-house software. In the short-latency afferent inhibition paradigm effects on the MEP of the factors ‘unconditioned MEP size’, ‘interstimulus interval’, ‘nerv’ and ‘muscle’ and their interactions were evaluated using a linear mixed model taking into account the correlations between multiple measurements within a participant. Compound symmetry was used as working correlation matrix. The final model included only significant factors and interactions. For these, pairwise differences of factor and interaction outcomes were evaluated. We also used repeated measures ANOVA to examine the effects of rTMS on MEP size.

To examine inhibition, or facilitation, at the intensity where the effects were largest we combined two inhibitory ISIs (N20, N20+2) to give one single value for inhibition, and two facilitatory ISIs (N20+12, N20+14) to result in one single value for facilitation. We used ANOVA to examine the effects statistically. Coefficients of variation were calculated on the single values for inhibition, or facilitation, as the ratio of the standard deviation to the mean. A statistical difference in the ANOVAs was followed by a post-hoc paired t-test analysis. Mauchly’s test was used to test for sphericity in the repeated measures ANOVAs, and the Greenhouse-Geisser correction was applied to the DFs if necessary. Statistical significance levels were set to p=0.05. All statistical analyses were performed using SAS version 9.2 or SPSS 16 for Windows software package.

1.4 Results

1.4.1 Experiment 1: Short-latency sensory afferent inhibition: The effect of recording site and conditioning stimulus intensity

Resting motor threshold for eliciting an MEP in FDI was 35.7% of stimulator output (SD 4.9, n=17). The threshold for electrical stimulation of the median nerve were 3.9mA (mean sensory threshold), and 6.9mA (mean motor threshold). When stimulating the ulnar nerve, the thresholds were 5.3mA (mean sensory threshold) and 6.5mA (mean motor threshold). The mean N20 SEP latency was 19.12ms (SD 0.94) for the median nerve and 19.93ms (SD 1.1) for the ulnar nerve. The effects of conditioning the motor cortex with electrical stimulation of median or ulnar nerve differed between individuals. At motor threshold conditioning intensity the coefficient of variation of SAI ranged from about 25 to 50% (table 1). Between two sessions the effects within the same participant were more stable ranging from about 12 to 30% (table 1).

At above motor threshold as the conventional intensity of median nerve stimulation, conditioning the MEP influenced its size when recorded from either FDI or APB (main effect of 'ISI' $F_{9,144}$=18.9, p<0.0001, Figure 1A). Overall, MEP size, and the response to conditioning differed between the two recording sites (main effect of 'muscle', $F_{1,16}$=7.54, p=0.0143) and between ulnar and median nerve stimulation ($F_{1,16}$=49.1, p<0.0001) with a different response at the recording site depending on the nerve stimulated (interaction 'nerve'*'muscle', $F_{1,16}$=12.49, p=0.0028, Figure 1A, B). Pairwise comparisons revealed a difference between APB and FDI when stimulating the median nerve (p=0.0005), and a difference between median and ulnar nerve when recording form APB (p<0.0001). The response in FDI was similar after stimulation of median or ulnar nerve, as was the response to ulnar nerve stimulation in APB and FDI (Figure 1B). Pairwise comparisons of the effect 'ISI'*'nerve' showed that the main difference was an increase in MEP size in APB, but not FDI, at ISIs of N20+8 (p=0.0007), N20+10 (p=0.0003), N20+12 (p<0.0001), N20+14 (p<0.0001), and N20+16 (p<0.0001), whereas at all other ISIs the decrease in MEP size was similar. While

the unconditioned MEP size differed between APB and FDI ($F_{1,623}$=578.32, p<0.0001) unconditioned MEP size had no statistical influence on the differential response to conditioning of ulnar, or median, nerve at any of the ISIs when recording from FDI or APB (unconditioned MEP size as covariate in the linear mixed model).

We then collapsed two inhibitory ISIs (N20+2, N20+4) to give one single value for inhibition, and two facilitatory ISIs (N20+12, N20+14) to result in one single value for facilitation. At motor threshold intensity where effects were most pronounced MEPs were smaller at inhibitory ISIs with median nerve or ulnar nerve stimulation whether recorded from the FDI or APB (Figure 1C). Only conditioning with median nerve stimulation and recording from APB induced clear facilitation (ANOVA, interaction 'stimulation site'*ISI', $F_{1,16}$=38.9, p<0.0001, Figure 1C).

We next assessed the effect of the intensity of the conditioning stimulus on the size of the MEP. Unconditioned MEPs were similar in FDI with median nerve conditioning (1.3mV (SD 0.54) at sensory threshold, 1.2mV (0.55) at sensory-motor, and 1.27 (0.48) at motor threshold); APB amplitudes were smaller (0.75mV (0.44), 0.76mV (0.55), 0.8mV (0.53)) at all intensities. With ulnar nerve conditioning the unconditioned MEPs were also similar at both intensities in FDI (1.31mV (0.66) and 1.25mV (0.4), or APB (0.87mV (0.58) and 0.93mV (0.61)). When the intensity of median nerve stimulation was raised the amount of inhibition increased (repeated measures ANOVA, main effect of intensity, $F_{1.4,29.9}$=6.3, p=0.013, Figure 2A and B). There was an increase in the amount of inhibition in FDI and APB; however, the response differed with more inhibition in FDI than in APB (repeated measures ANOVA on data in Figure 1A and B, interaction 'site'*'intensity', $F_{1.8}$, 29.5=3.7, p=0.039). Ulnar nerve stimulation also induced inhibition the amount of which increased with the stimulation intensity (repeated measures ANOVA; $F_{1,16}$=12.6, p=0.003, Figure 2C and D) but was not different between APB and FDI. Facilitation was observed only when the median nerve was stimulated and the MEP recorded from APB (main effect of 'site', $F_{1,16}$=10.2, p=0.006). The amount of facilitation increased in APB with increasing stimulation intensity; however, when

including all stimulation intensities and both recording sites, this was not significant. Facilitation was not observed when conditioning with ulnar nerve stimulation.

1.4.2 Experiment 2: Short-latency sensory afferent inhibition after motor cortex 1Hz repetitive transcranial magnetic stimulation

With the Magstim Rapid stimulator, the motor threshold was 51.6% of stimulator output (SD 5.3). The inhibitory rTMS protocol (1500 stimuli, 95% motor threshold at 1Hz) reduced MEP size in FDI, or APB, by about 20-30% for at least 20 minutes after rTMS (FDI: repeated measures ANOVA, $F_{2,30}$=14,66, p<0.0001; APB: $F_{2,30}$=16.45, p<0.0001; *p=0.001, post-hoc t-test unconditioned versus immediately after rTMS, **p=0.001, post-hoc t-test unconditioned versus 20 minutes after rTMS, Figure 3A, B). RTMS had, however, no effect on the amount of inhibition or facilitation in the sensory afferent paradigm after median nerve conditioning and FDI, or APB, MEP recording (Figure 3B). MEP amplitudes in FDI were slightly larger than in APB before (1.32mV (SD 0.55) versus 0.93mV (SD 0.38) and after rTMS (1.17mV (SD 0.43) versus 0.93mV (SD 0.83). Unconditioned MEP amplitudes in the SAI paradigm were similar before and after rTMS suggesting the adjustment of the TMS stimulation intensity was appropriate. MEP latencies were also similar before and after rTMS (table 2), as were long-loop reflex latencies (table 2), the area under the curve of LLR2 and the cortical relay times (table 2).

1.5 Discussion

In the present study we evaluated the influence on SAI of conditioning stimulus intensity, conditioning site and recording site. Our data show that 1) the variability between individuals is high but within the same participant the variability between different sessions is much lower; 2) median nerve or ulnar nerve stimulation induces inhibition in homotopic and heterotopic muscles; 3) only in APB as homotopic median nerve muscle there is facilitation; 4) stimulus intensity determines the amount of inhibition or facilitation. Inhibitory motor cortex rTMS reduced excitability of the motor cortex but had no effect on SAI, long-loop reflexes or cortical relay time.

1.5.1 Short-latency sensory afferent inhibition: stimulus intensity and recording site

Conventionally, when stimulating a mixed sensory-motor nerve to condition the motor cortex an intensity just above motor threshold of that nerve is used [3]. As our data confirm this leads to robust inhibition at ISIs around the N20 latency of the SEP [11]. In addition, at ISIs 8-16ms longer than the individual N20 latency stimulating the median nerve induces facilitation when recording from the APB [11]. We extended these observations by showing that the amount of inhibition and facilitation depends on the intensity of the conditioning stimulus as had been demonstrated previously in a small group of healthy individuals (Di Lazzaro). There was an effect on MEP size at just above sensory threshold, the lowest stimulation intensity; however, the amount of inhibition, or facilitation, increased to its maximum when we raised the conditioning stimulus intensity to just above motor threshold. Stimulating the ulnar nerve, sensory threshold and motor threshold were too close together to allow an intermediate stimulation intensity. In the median nerve, an intensity of conditioning stimulus between sensory and motor thresholds resulted in intermediate inhibition or facilitation. At sensory thresholds of the median, or ulnar, nerve the stimulation of sensory and motor afferents of these mixed nerves has a similar effect on motor cortex excitability than cutaneous, i.e. sensory afferents only, conditioning using ring electrodes [3, 9, 10]. The increase of the amount of inhibition, or facilitation, with increasing peripheral nerve conditioning stimulation

intensity suggests that stimulating more sensory fibres can enhance the effect of peripheral nerve conditioning on motor cortex excitability. There is good evidence to suggest that SAI occurs in the cortex [3]; the timings between peripheral nerve conditioning and the inhibitory effect on motor output indicate that this involves at least one synapse. An increase of the stimulation intensity at the peripheral nerve may thus, at a cortical level, secondarily lead to synaptic release of inhibitory, or facilitatory, neurotransmitters.

We next looked at differences depending on the recording site. The experimental set-up allowed assessing a conditioning effect in an anatomically homotopic muscle – first dorsal interosseus for ulnar nerve, and abductor pollicis brevis for median nerve – while also recording the response to stimulation in an anatomically heterotopic muscle – first dorsal interosseus for median nerve, and abductor pollicis brevis for ulnar nerve. Stimulating either peripheral nerve at ISIs around the N20 latency of the SEP reduced motor cortex excitability in homotopic and heterotopic muscles. This suggests that the inhibitory effects of peripheral nerve conditioning on the motor cortex are not confined to the representation of homotopic muscles in the motor cortex but extend to adjacent muscle representations. Consistent with this notion we observed facilitation in the anatomically homotopic median nerve muscle at ISIs about 10-20ms later than the N20 latency, confirming previous observations [11]. We did not see such a facilitatory effect in the heterotopic FDI muscle after median nerve stimulation suggesting this effect is specific to the homotopic muscle. A different type of somatosensory input, vibration, enhanced the excitability in circuits controlling motor output in those small hand muscles that were vibrated and attenuated it in those that were not [19]. Together with our data from median nerve sensory input this would suggest that somatosensory inputs shape the cortical motor command in a functionally relevant way with opposite effects in agonist and antagonist muscles. If so, we would have expected to observe similar effects with ulnar nerve stimulation. However, after ulnar nerve conditioning there was no facilitatory effect of the MEP in either muscle. It is not clear why a facilitatory effect should be specific to the homotopic motor cortex representations of a median nerve muscle only. It is of note, though, that at higher ulnar nerve stimulation intensities the amount of inhibition increased at

early ISIs while it decreased at later ISIs. We speculate that there is a mix of effects at late ISIs such that when stimulus intensity increases there is more facilitation that then reduces the apparent amount of inhibition. Another possible confounding factor might be cutaneous afferents that are activated when stimulating the peripheral nerve through the skin. Cutaneous digital nerve stimulation can also modulate the excitability of the motor output [3, 9, 10]. The inhibitory effects of cutaneous digital nerve stimulation were more pronounced in the muscles near the stimulated finger, and these were called homotopic independent of the anatomical relation of stimulated skin and muscle that was recorded from [9, 10]. These effects were observed at ISIs of up to 40ms [9, 10]; at similar ISIs we observed no inhibition with peripheral nerve stimulation at above its sensory threshold and facilitation when stimulating at above its motor threshold. Strong facilitation in the muscle nearest the stimulation site at the wrist (APB) does not implicate a major contribution of inhibitory effects from cutaneous afferents. This suggests that the mechanisms differ through which cutaneous stimulation, or direct electrical peripheral nerve stimulation, influence motor cortex excitability. Interestingly, Quartarone and colleagues found homotopic effects when using a 5Hz rapid-rate paired associative stimulation protocol, a protocol that resembles repetitive SAI [20]. These authors observed facilitatory effects at an ISI of 25ms, which in SAI reduces motor cortex excitability. These directly opposite effects suggest that despite their resemblance rapid-rate paired associative stimulation differs quite substantially from SAI.

1.5.2 The effect of 1Hz repetitive transcranial magnetic stimulation

In the second experiment we examined the effects on SAI and long-loop reflexes of inhibitory rTMS. As previously shown, rTMS at 1Hz and an intensity of 95% resting motor threshold reduced MEP amplitude in the relaxed hand for at least 20 minutes [15, 21-23]. However, rTMS had no effect on SAI, or facilitation. In addition, rTMS did not change the latency and size of the long-loop reflexes or the cortical relay time from the arrival of the sensory afferent volley in the somatosensory cortex to the MEP produced from the discharge of motoneurones in the motor cortex. This suggests that the inhibitory effects of rTMS were

confined to those neurones responsible for the MEP without any effects on the excitability of the somatosensory cortex and the elements that make up the transcortical relay from somatosensory to motor cortex [12, 13]). Consistent with this interpretation Enomoto and colleagues showed that a similar inhibitory rTMS protocol had no effect on the N20 latency of the SEP which is thought to reflect the arrival of the sensory input at the somatosensory cortex [14]. Bäumer and colleagues observed that inhibitory rTMS given to the somatosensory cortex, or the motor cortex, also had no effect on SAI in healthy individuals [24]. However, in their patients with writer's cramp, a focal dystonia with abnormal somatosensory systems, rTMS to the somatosensory system reduced SAI [24]. Thus, the somatosensory systems of healthy individuals, but not patients with focal dystonia, may compensate sufficiently for the effects of rTMS. Another explanation is that the state of the muscle, active or relaxed, may influence the effects of rTMS [25]. Some of the paradigms - SEP, long latency reflexes - used to assess the effects of rTMS on the somatosensory cortex or sensori-motor integration require muscle activation. We show that rTMS has no effect even when we use short-latency sensory afferent inhibition where sensory inputs are given with muscles at rest. Based on these observations we conclude that pre-activation cannot be the only explanation of why we saw no effect of inhibitory rTMS beyond the MEP.

In conclusion our data indicate that it is possible to assess input-output curves in the SAI pathway. This may complement the investigation of cortical sensori-motor integration in healthy individuals but also disorders where sensori-motor integration is abnormal, e.g. dystonia. In addition, we demonstrate that the inhibitory effects of preconditioning the motor cortex with peripheral nerve stimulation are not limited to anatomically homotopic muscles. Finally, we show that the effects of an inhibitory rTMS protocol are specific to the motor output involved in the stimulation but do not modulate the interaction of sensory inputs with the motor cortex.

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1.8 Legends

Table 1. Between subject and between session coefficient of variability

	Recording from	Ulnar nerve Inhibition	facilitation	Median nerve Inhibition	facilitation
Between subject	FDI	37.5	24.3	49.7	27.5
Between subject	APB	38.3	22.5	46.8	50.5
Between session	FDI	n.a.	n.a.	12.7	21.9
Between session	APB	n.a.	n.a.	14.8	31.7

Table 1: Between subject and between session coefficient of variability of short latency sensory-afferent inhibition with motor threshold stimulation intensity. Two inhibitory ISIs (N20, N20+2) were combined to give one single value for inhibition, and two facilitatory ISIs (N20+12, N20+14) to result in one single value for facilitation. Coefficients of variability were calculated as the ratio of the standard deviation to the mean. Abbreviations: FDI: first dorsal interosseus. APB: abductor pollicis brevis. N.a. not available. ISI: inter-stimulus interval

Figure 1

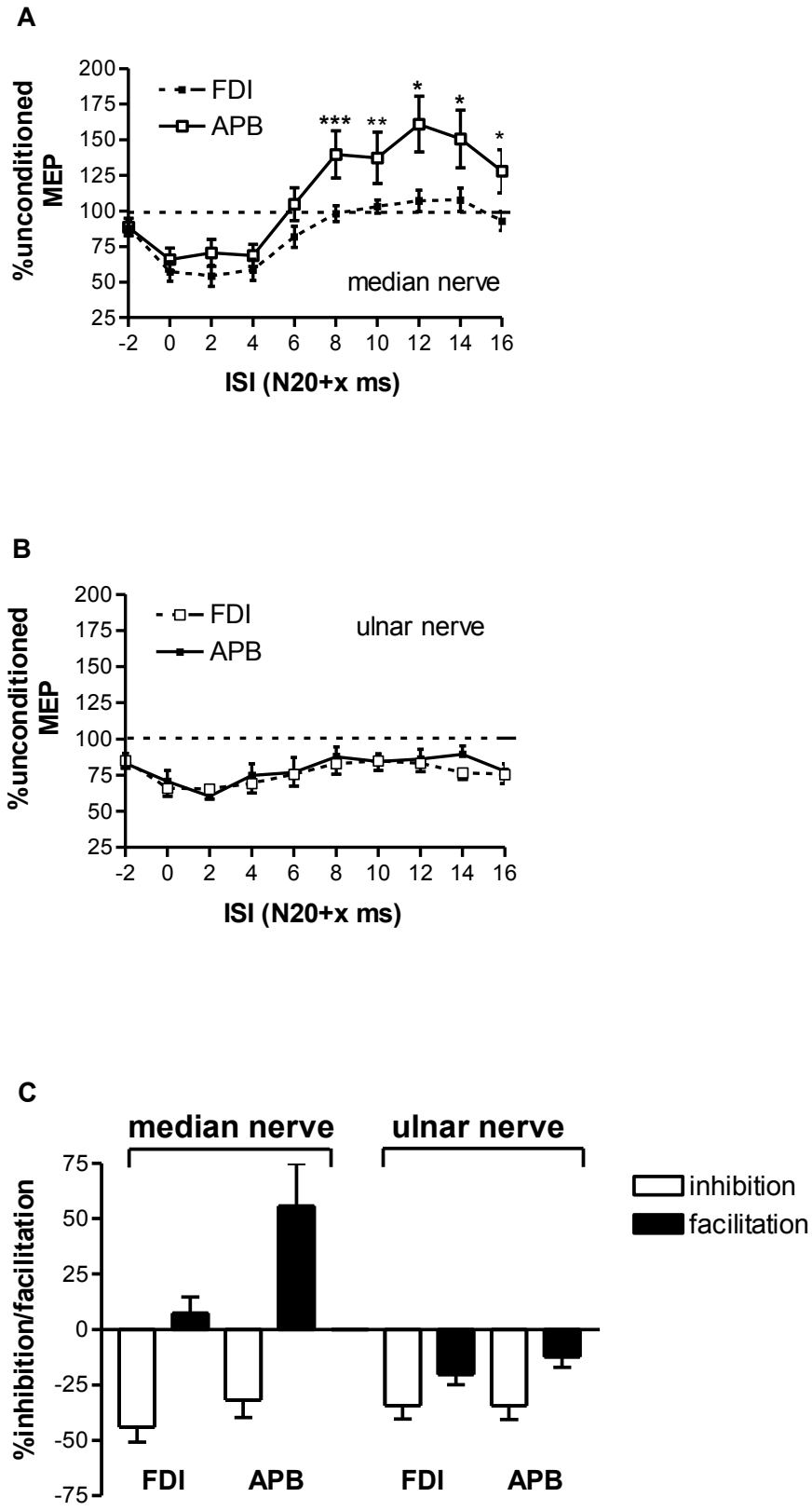
A
%unconditioned MEP
FDI
APB
median nerve
ISI (N20+x ms)
B
%unconditioned MEP
FDI
APB
ulnar nerve
ISI (N20+x ms)
C
median nerve
ulnar nerve
%inhibition/facilitation
inhibition
facilitation
FDI
APB
FDI
APB

Figure 1. At above motor threshold, rTMS conditioning changed MEP size when recorded from either FDI or APB (mixed linear model, main effect of 'muscle', $F_{1,16}$=7.54, p=0.0143) and between ulnar and median nerve stimulation (mixed linear model, $F_{1,16}$=49.1, p<0.0001) with a different response at the recording site depending on the nerve stimulated (interaction 'nerve'*'muscle', $F_{1,16}$=12.49, p=0.0028, A, B). Pairwise comparisons revealed a difference between APB and FDI when stimulating the median nerve (p=0.0005) and a difference between median and ulnar nerve when recording form APB (p<0.0001). The response in FDI was similar after stimulation of median or ulnar nerve, as was the response to ulnar nerve stimulation in APB and FDI (B). Pairwise comparisons of the effect 'ISI'*'nerve' showed that the main difference was an increase in MEP size in APB, but not FDI, at ISIs of N20+8 (***p=0.0007), N20+10 (**p=0.0003), N20+12 (*p<0.0001), N20+14 (*p<0.0001), and N20+16 (*p<0.0001), whereas at all other ISIs the decrease in MEP size was similar. **C.** To illustrate these effects better we collapsed two inhibitory ISIs (N20, N20+2) to give one single value for inhibition, and two facilitatory ISIs (N20+12, N20+14) to result in one single value for facilitation. MEPs were smaller at inhibitory ISIs with median nerve or ulnar nerve stimulation whether recorded from the FDI or APB. Only conditioning with median nerve stimulation and recording from APB induced clear facilitation (ANOVA, interaction 'stimulation site'*ISI', $F_{1,16}$=38.9, p<0.0001). Abbreviations: FDI: first dorsal interosseus. APB: abductor pollicis brevis. ISI: inter-stimulus interval. MEP: motor evoked potential.

Figure 2

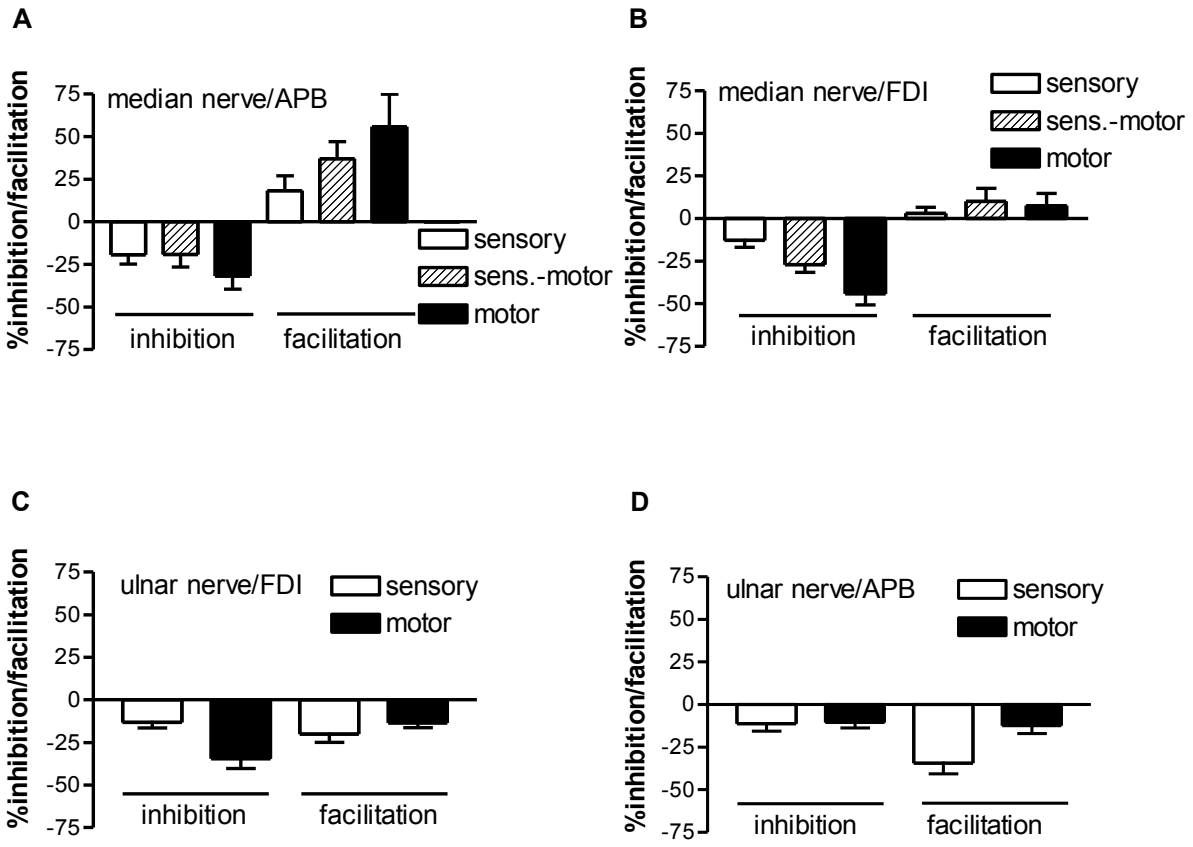

Figure 2. Effect of the intensity of the conditioning stimulus on the size of the MEP. **A, B.** Median nerve stimulation. The amount of inhibition increased with increasing stimulation intensity in FDI and APB (repeated measures ANOVA, main effect of intensity, $F_{1.4,29.9}$=6.3, p=0.013). There was more inhibition in FDI than in APB (repeated measures ANOVA, interaction 'site'*'intensity', $F_{1.8}$, 29.5=3.7, p=0.039). Facilitation was observed when the MEP was recorded from APB (main effect of 'site', $F_{1,16}$=10.2, p=0.006) but not FDI. **C, D.** Ulnar nerve stimulation. The amount of inhibition increased with the stimulation intensity (repeated measures ANOVA; $F_{1,16}$=12.6, p=0.003, Figure 2C and D) but was not different between APB and FDI. Facilitation was not observed. Abbreviations: FDI: first dorsal interosseus. APB: abductor pollicis brevis. MEP: motor evoked potential.

Figure 3.

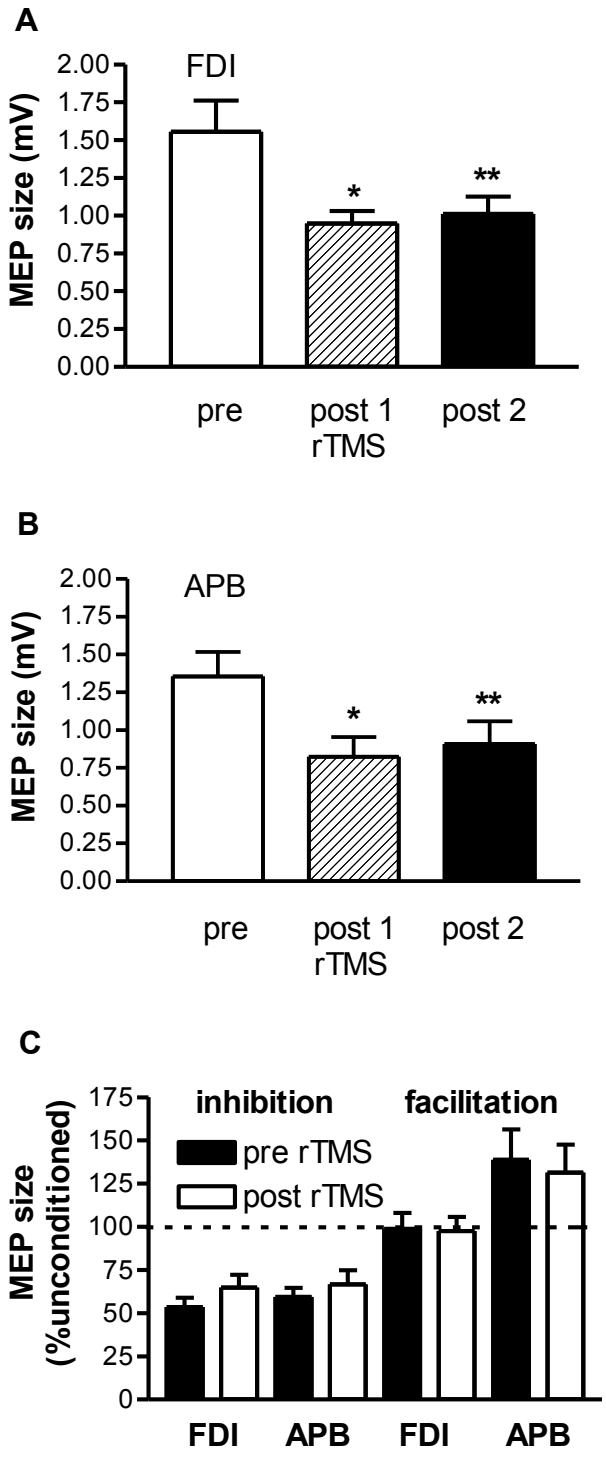

Figure 3. Effects of repetitive transcranial magnetic stimulation (rTMS). A, B. The inhibitory rTMS protocol (1500 stimuli, 95% motor threshold at 1Hz) reduced MEP size by about 20-30% for at least 20 minutes after rTMS (repeated measures ANOVA, FDI: $F_{2,30}$=14,66, p<0.0001; APB: $F_{2,30}$=16.45, p<0.0001; *p=0.001, post-hoc t-test unconditioned versus

immediately after rTMS, **p=0.001, post-hoc t-test unconditioned versus 20 minutes after rTMS). C. There was no effect on the amount of inhibition or facilitation in the short-latency sensory afferent paradigm after median nerve conditioning and FDI, or APB, MEP recording. Abbreviations: FDI: first dorsal interosseus. APB: abductor pollicis brevis. MEP: motor evoked potential.

2 Summary

Short-latency sensory afferent inhibition: conditioning stimulus intensity, recording site and effects of 1Hz repetitive TMS

M Fischer, M Orth MD PhD

Department of Neurology, Universitätsklinikum Ulm, Oberer Eselsberg 45/1, 89081 Ulm, Germany

2.1 Introduction

Sensori-motor integration describes the process by which the sensory and motor systems communicate and coordinate their activities. It involves the reception of a stimulus and its transmission to the central nervous system where the stimulus is interpreted. In case of a motor response this involves the transmission of impulses along corticospinal motoneurones to a group of muscles, which elicits an appropriate movement. Sensori-motor integration is an integral part of day-to-day life.

The regions of the brain responsible for receiving sensory information (thalamus, primary somatosensory cortex, postcentral gyrus, primary gustatory area, primary auditory cortex, primary visual cortex, olfactory cortex) and those executing movements (primary motor cortex, premotor cortex, supplementary motor area, posterior parietal cortex and several subcortical brain regions) therefore have to interact. Recent data also has shown that these parts of the brain are not only specified for one single sensory modality, but responsible for multiple different sensory inputs coming from the environment [1-3]. Examples for abnormal sensorimotor integration are some movement disorders. Pathophysiologically, the common view is that patients suffering from movement disorders have a dysfunction of the basal ganglia – motor cortex circuits. Anatomically, the basal ganglia receive topographically organized inputs from nearly all cortical regions. Later on, these inputs are being sent to their targets, e.g. cortical areas that are involved in cognition. The basal ganglia may, in addition to other functions, serve as a gate that filters sensory inputs so that information from the environment relevant for guiding movements reaches the motor areas. If something goes wrong within these circuits subserving sensory motor integration the results may be abnormal processing of motor programs. This may contribute to the pathophysiology of

movement disorders. It is thought that people having particularly movement disorders are not able to use sensory information properly for assisting motor actions. Examples are Parkinson´s disease and dystonia. For instance, in Parkinson´s Disease the reliance of patients on visual information during movement actions may indicate that there is a defect in proprioception. This disturbance of proprioceptive regulation that is probably related to the occurance of abnormal muscle stretch reflexes, could be important for some of the symptoms in Parkinsons´s disease such as hypometria or bradykinesia [4].

On the other hand especially dystonia is a good example for abnormal sensorimotor integration, where sensory manipulation can modify abnormal movements. The most common phenomenon is the “geste antagonistique”, a trick in which the patient touches a body part close to the part with abnormal posture (often the neck and face), thus producing special tactile or proprioceptive sensory input. As a result a dystonic posture may improve [4].

There are different experimental ways of investigating sensorimotor integration. One option is magnetic resonance imaging (MRI) to investigate the neural representation of sensorimotor integration. Sasaki et al. used functional MRI to investigate the neural basis of sensory afferents in the primary sensorimotor cortex [5]. Whenever a ball was rotated either by the investigator’s hand or automatically, the signal in the sensorimotor cortex was enhanced. Many different sensory stimuli were used to investigate sensorimotor integration with functional MRI, such as “pain” in the study of Morrison et al, where distinct sensorimotor subregions emerged in the brains of respondants by “feeling” other painful actions [6]. In musicians with embouchure (muscles of the lips) dystonia, there was a significant “increased activation of somatotopic face representations within the bilateral primary sensorimotor cortex” [7]. In addition, bilaterally the premotor cortex was activated significantly during buzzing an instrument-specific mouthpiece [7]. This led to the conclusion that this could reflect deficient subcortical and intracortical inhibition or abnormal sensorimotor integration and reorganization. On the other hand Thomalla and colleagues have shown with structural MRI that in patients with Gilles de la Tourette Syndrome the sensorimotor cortex was thinner than in healthy controls [8-9].

Transcranial magnetic stimulation methods (TMS) can examine the effect of afferent sensory inputs on the excitability of human motor cortex. Motor evoked potentials (MEPs) are modulated by a preceding electrical stimulus to mixed nerves or cutaneous nerves.

A paradigm frequently used is short-latency sensory afferent inhibition (SAI). Here an electrical stimulus given to a mixed nerve precedes the TMS pulse. At inter-stimulus intervals slightly longer than the N20 component of somatosensory evoked potentials (SEPs) this reduces the size of the MEP. In addition, at longer inter-stimulus intervals between conditioning stimuli and TMS shock to the motor cortex the MEP size increases.

We extended the investigation of SAI in three ways: 1) we investigate coefficients of variation between sessions and between subjects; 2) we examined the effects of different intensities of mixed nerve stimulation on MEP size; 3) we stimulated median and ulnar nerve, and recorded simultaneously from first dorsal interosseus (FDI) and abductor pollicis brevis (APB) muscles to assess the differential effects on anatomically homotopic, or heterotopic, muscles.

In a second experiment we stimulated the motor cortex using an inhibitory repetitive TMS protocol to examine the effects on MEP size, SAI, long-loop reflexes and the relay time from somatosensory cortex to motor cortex. Our hypothesis was that the inhibitory effects of rTMS conditioning extend beyond the motor cortex and the somatosensory cortex to involve the integration of sensory stimuli with motor output.

2.2 Material and Methods

2.2.1 Participants

Seventeen healthy right handed Caucasian volunteers were studied (mean age 24, range 23-27, 5 women). Subjects were asked to refrain from coffeine-containing beverages on the day of the experiment. All participants took part in experiment one; 16 participants took part in the second experiment. Participants gave informed written consent, and the local ethics committee approved the study protocol.

2.2.2 Electromyography recordings

Surface electromyograms (EMG) were recorded from the right FDI and the right APB muscle. The EMG signal was amplified and analogue filtered (30Hz to 1kHz).

2.2.3 Transcranial Magnetic Stimulation

Participants were seated in a comfortable chair. They were asked to relax as much as possible. Magnetic stimuli were given with a hand-held figure-of-eight coil (outer winding diameter 9cm). The optimal spot for right FDI and APB stimulation was marked with a felt pen.

RTMS (1Hz, 1800 stimuli to the left motor cortex at 95% resting motor threshold) was applied using a Magstim Rapid (Magstim Co., Whitland, Dyfed, UK) which generates a biphasic (posterior-anterior/anterior posterior) current flow in the brain.

2.2.4 Motor thresholds

Resting motor threshold (RMT) was defined as the minimum intensity needed to evoke an MEP of >50μV in 5 out of 10 consecutive trials in the relaxed FDI. Active motor threshold (AMT) was defined as the minimum intensity (in % of maximum stimulator output) needed to evoke a MEP of >200μV in 5 out of 10 trials in the tonically active FDI.

2.2.5 Somatosensory evoked potentials

After stimulation of the median or ulnar nerve with surface electrodes at threshold intensities for motor stimulation, somatosensory evoked potentials were recorded with a needle electrode over

the somatosensory cortex (2cm posterior of C3 in the international EEG 10-20 system) referenced against the opposite ear lobe. Stimulation was given at a frequency of 3 Hz; a total of 300 stimuli were averaged.

2.2.6 Short latency afferent inhibition (SAI) by somatosensory input from the median, or ulnar, nerve

SAI of the motor cortex was examined as previously described. The electrical stimulus to the median nerve was delivered at three different intensities: above sensory threshold, just above the threshold to elicit a visible contraction in the thenar muscles, and at an intensity between sensory and motor threshold. To the ulnar nerve, electrical stimulation was delivered above sensory threshold and above motor threshold. Since the difference between sensory and motor threshold was smaller than for the median nerve we did not use a third intensity between sensory and motor threshold. Stimulation preceded the TMS pulse in relation to the N20 component of sensory evoked potentials (N20, N20+2, N20+4, N20+6, N20+8, N20+10, N20+12, N20+14, N20+16). Twenty trials of the MEP elicited by TMS alone and 10 trials of conditioned MEPs for each inter-stimulus interval (ISI) were collected. The amplitude of the MEP was measured with in-house software. The intensity of the TMS pulse was adjusted to result in a MEP of between 0.5 and 1mV in amplitude. The intensity was adjusted again following rTMS so that unconditioned MEP amplitude was similar before and after rTMS. The average amplitude of the conditioned MEP was expressed in percent of the average amplitude of the un-conditioned MEP alone.

2.2.7 Long-latency reflexes

Reflexes were elicited in the contracted right abductor pollicis brevis muscle by electrical stimulation of the median nerve at the wrist. Subjects sat with their pronated forearm supported before them on a table and contracted the APB muscle isometrically by abducting the thumb against a force transducer with reference to a visual display before them. The median nerve was stimulated at motor threshold intensity. We visually inspected the reflexes following electrical nerve stimulation on average records of full-wave rectified EMG activity. Then we determined the end of the short latency and beginning of the long latency reflex when the average surface rectified EMG

increased abruptly at a latency of between 45 and 55 ms. First, we estimated the duration of the short and long latency components of the EMG responses. Then, we calculated the integral of the rectified EMG activity as the size of the reflexes.

2.2.8 Data analysis

Peak to peak amplitude of MEP was measured with in-house software. In the short-latency afferent inhibition paradigm effects on the MEP of the factors ‘unconditioned MEP size’, ‘interstimulus interval’, ‘nerv’ and ‘muscle’ and their interactions were evaluated using a linear mixed model taking into account the correlations between multiple measurements within a participant. The final model included only significant factors and interactions. For these, pairwise differences of factor and interaction outcomes were evaluated. We also used repeated measures ANOVA to examine the effects of rTMS on MEP size.

To examine inhibition, or facilitation, at the intensity where the effects were largest we combined two inhibitory ISIs (N20, N20+2) to give one single value for inhibition, and two facilitatory ISIs (N20+12, N20+14) to result in one single value for facilitation.

A statistical difference in the ANOVAs was followed by a post-hoc paired t-test analysis. Mauchly’s test was used to test for sphericity in the repeated measures ANOVAs, and the Greenhouse-Geisser correction was applied to the DFs if necessary. Statistical significance levels were set to p=0.05. All statistical analyses were performed using SAS version 9.2 or SPSS 16 for Windows software package.

2.3 Results

2.3.1 Experiment 1: Short-latency sensory afferent inhibition: The effect of recording site and conditioning stimulus intensity

Resting motor threshold for eliciting an MEP in FDI was 35.7% of stimulator output. The thresholds for electrical stimulation of the median nerve were 3.9mA (mean sensory threshold), and 6.9mA (mean motor threshold). When stimulating the ulnar nerve, the thresholds were 5.3mA (mean sensory threshold) and 6.5mA (mean motor threshold). The mean N20 SEP latency was 19.12ms for the median nerve and 19.93ms for the ulnar nerve. The effects of conditioning the motor cortex with electrical stimulation of median or ulnar nerve differed between individuals. At motor threshold conditioning intensity the coefficient of variation of SAI ranged from about 25 to 50%. Between two sessions the effects within the same participant were more stable ranging from about 12 to 30%.

At above motor threshold of median nerve stimulation, conditioning the MEP influenced its size when recorded from either FDI or APB. Overall, MEP size, and the response to conditioning stimulation differed between the two recording sites and between ulnar and median nerve stimulation with a different response at the recording site. Pairwise comparisons revealed a difference between APB and FDI when stimulating the median nerve, and a difference between median and ulnar nerve when recording from APB. The response in FDI was similar after stimulation of median or ulnar nerve, as was the response to ulnar nerve stimulation in APB and FDI. Pairwise comparisons of the effect 'ISI'*'nerve' showed that the main difference was an increase in MEP size in APB, but not FDI, at ISIs of N20+8, N20+10, N20+12, N20+14, and N20+16, whereas at all other ISIs the decrease in MEP size was similar. While the unconditioned MEP size differed between APB and FDI, unconditioned MEP size had no statistical influence on the differential response to conditioning of ulnar, or median, nerve at any of the ISIs when recording from FDI or APB. We then collapsed two inhibitory ISIs (N20+2, N20+4) to give one single value for inhibition, and two facilitatory ISIs (N20+12, N20+14) to result in one single value for facilitation. At motor threshold intensity where effects were most pronounced MEPs were smaller at inhibitory ISIs with median nerve or ulnar nerve stimulation whether recorded from the FDI or APB. Only conditioning with median nerve stimulation and recording from APB induced clear facilitation.

We next assessed the effect of the intensity of the conditioning stimulus on the size of the MEP. Unconditioned MEPs were similar in FDI with median nerve conditioning; APB amplitudes were smaller at all intensities. With ulnar nerve conditioning the unconditioned MEPs were also similar at both intensities in FDI or APB. When the intensity of median nerve stimulation was raised the amount of inhibition increased. There was an increase in the amount of inhibition in FDI and APB; however, the response differed with more inhibition in FDI than in APB. Ulnar nerve stimulation also induced inhibition the amount of which increased with the stimulation intensity but was not different between APB and FDI. Facilitation was observed only when the median nerve was stimulated and the MEP recorded from APB. The amount of facilitation increased in APB with increasing stimulation intensity; however, when including all stimulation intensities and both recording sites, this was not significant. Facilitation was not observed when conditioning with ulnar nerve stimulation.

2.3.2 Experiment 2: Short-latency sensory afferent inhibition after motor cortex 1Hz repetitive transcranial magnetic stimulation

The motor threshold was 51.6% of stimulator output. The inhibitory rTMS protocol (1500 stimuli, 95% motor threshold at 1Hz) reduced MEP size in FDI, or APB, by about 20-30% for at least 20 minutes after rTMS. RTMS had, however, no effect on the amount of inhibition or facilitation in the sensory afferent paradigm after median nerve conditioning and FDI, or APB, MEP recording. MEP amplitudes in FDI were slightly larger than in APB before and after rTMS. Unconditioned MEP amplitudes in the SAI paradigm were similar before and after rTMS suggesting the adjustment of the TMS stimulation intensity was appropriate. MEP latencies were also similar before and after rTMS, as were long-loop reflex latencies, the area under the curve of LLR2 and the cortical relay times.

2.4 Discussion

We evaluated the influence on SAI of conditioning stimulus intensity, conditioning site and recording site. Our data show that 1) the variability between individuals is high but within the same participant the variability between different sessions is much lower; 2) median nerve or ulnar nerve stimulation induces inhibition in homotopic and heterotopic muscles; 3) only in APB as homotopic median nerve muscle there is facilitation; 4) stimulus intensity determines the amount of inhibition or facilitation. Inhibitory motor cortex rTMS reduced excitability of the motor cortex but had no effect on SAI, long-loop reflexes or cortical relay time.

2.4.1 Short-latency sensory afferent inhibition: stimulus intensity and recording site

Our data confirm that a peripheral nerve stimulation intensity above motor threshold leads to robust inhibition at ISIs around the N20 latency of the SEP. In addition, at ISIs 8-16ms longer than the individual N20 latency, stimulating the median nerve induces facilitation when recording from the APB. We extended these observations by showing that the amount of inhibition and facilitation depends on the intensity of the conditioning stimulus. The amount of inhibition, or facilitation, increased to its maximum when we raised the conditioning stimulus intensity to just above motor threshold. In the median nerve, an intensity of conditioning stimulus between sensory and motor thresholds resulted in intermediate inhibition or facilitation. The increase of the amount of inhibition, or facilitation, with increasing peripheral nerve conditioning stimulation intensity suggests that stimulating more sensory fibres can enhance the effect of peripheral nerve conditioning on motor cortex excitability. There is good evidence to suggest that SAI occurs in the cortex; the timings between peripheral nerve conditioning and the inhibitory effect on motor output indicate that this involves at least one synapse. An increase of the stimulation intensity at the peripheral nerve may thus secondarily lead to synaptic release of inhibitory, or facilitatory, neurotransmitters.

We next looked at differences depending on the recording site. The experimental set-up allowed assessing a conditioning effect in an anatomically homotopic muscle – first dorsal interosseus for ulnar nerve, and abductor pollicis brevis for median nerve – while also recording the response to stimulation in an anatomically heterotopic muscle – first dorsal interosseus for median nerve, and abductor pollicis brevis for ulnar nerve. Stimulating either peripheral nerve at ISIs around the N20

latency of the SEP reduced motor cortex excitability in homotopic and heterotopic muscles. This suggests that the inhibitory effects of peripheral nerve conditioning on the motor cortex are not confined to the representation of homotopic muscles in the motor cortex. Consistent with this notion we observed facilitation in the anatomically homotopic median nerve muscle at ISIs about 10-20ms later than the N20 latency. We did not see such a facilitatory effect in the heterotopic FDI muscle suggesting this effect is specific to the homotopic muscle. A different type of somatosensory input, vibration, enhanced the excitability in circuits controlling motor output in those small hand muscles that were vibrated and attenuated it in those that were not. Together with our data from median nerve sensory input this would suggest that somatosensory inputs shape the cortical motor command in a functionally relevant way with opposite effects in agonist and antagonist muscles. If so, we would have expected to observe similar effects with ulnar nerve stimulation. However, after ulnar nerve conditioning there was no facilitatory effect of the MEP in either muscle. It is not clear why a facilitatory effect should be specific to the homotopic motor cortex representations of a median nerve muscle only. It is of note, though, that at higher ulnar nerve stimulation intensities the amount of inhibition increased at early ISIs while it decreased at later ISIs. We speculate that there is a mix of effects at late ISIs such that when stimulus intensity increases there is more facilitation that then reduces the apparent amount of inhibition. Another possible confounding factor might be cutaneous afferents that are activated when stimulating the peripheral nerve through the skin. Cutaneous digital nerve stimulation can also modulate the excitability of the motor output. The inhibitory effects of cutaneous digital nerve stimulation were more pronounced in the muscles near the stimulated finger, and these were called homotopic independent of the anatomical relation of stimulated skin and muscle that was recorded from. These effects were observed at ISIs of up to 40ms; at similar ISIs we observed no inhibition with peripheral nerve stimulation at above its sensory threshold and facilitation when stimulating at above its motor threshold. Strong facilitation in the muscle nearest the stimulation (APB) does not implicate a major contribution of inhibitory effects from cutaneous afferents. This suggests that the mechanisms differ through which cutaneous stimulation, or direct electrical peripheral nerve stimulation, influence motor cortex excitability.

2.4.2 The effect of 1Hz repetitive transcranial magnetic stimulation

As previously shown, rTMS at 1Hz and an intensity of 95% resting motor threshold reduced MEP amplitude in the relaxed hand for at least 20 minutes. However, rTMS had no effect on SAI, or facilitation. In addition, rTMS did not change the latency and size of the long-loop reflexes or the cortical relay time from the arrival of the sensory afferent volley in the somatosensory cortex to the motor cortex. This suggests that the inhibitory effects of rTMS were confined to those neurones responsible for the MEP without any effects on the excitability of the somatosensory cortex and the elements that make up the transcortical relay from somatosensory to motor cortex. Bäumer and colleagues observed that inhibitory rTMS given to the somatosensory cortex, or the motor cortex, also had no effect on SAI in healthy individuals. However, in their patients with writer's cramp, rTMS to the somatosensory system reduced SAI. Thus, the somatosensory systems of healthy individuals, but not patients with focal dystonia, may compensate sufficiently for the effects of rTMS. Another explanation is that the state of the muscle, active or relaxed, may influence the effects of rTMS. Some of the paradigms - SEP, long latency reflexes - used to assess the effects of rTMS on the somatosensory cortex or sensori-motor integration require muscle activation. We show that rTMS has no effect even when we use short-latency sensory afferent inhibition where sensory inputs are given with muscles at rest. Based on these observations we conclude that pre-activation cannot be the only explanation of why we saw no effect of inhibitory rTMS beyond the MEP.

In conclusion our data indicate that it is possible to assess input-output curves in the SAI pathway. This may complement the investigation of cortical sensori-motor integration in healthy individuals but also disorders where sensori-motor integration is abnormal, e.g. dystonia. In addition, we demonstrate that the inhibitory effects of preconditioning the motor cortex with peripheral nerve stimulation are not limited to anatomically homotopic muscles. Finally, we show that the effects of an inhibitory rTMS protocol are specific to the motor output but do not modulate the interaction of sensory inputs with the motor cortex.

Ridding and Rothwell also mentioned that the effects of rTMS are not focal but depend on the prior history of brain activity [10]. This is important with a view to the question whether rTMS offers therapeutic potential or not. As rTMS is able to influence human behaviour, rTMS could also boost

function in suboptimally functioning parts of the brain or reduce activity of parts that interfere with recovery, e.g. after brain injury or in chronic CNS diseases such as dystonia. Furthermore they posit that rTMS might be therapeutic by correcting an imbalance in function that has been caused by a disease – also when symptoms reoccur [11]. On the other hand rTMS to the injured brain is also being considered to accelerate normal adaptations in treatment after a brain injury, which means that there could be a faster restoration of normal brain function [11]. To date, rTMS has only been used in a larger number of patients with depression, with suggestions of effects also reported in Dystonia, Parkinson´s disease, Stroke and Neurogenic pain [11;13]. In Depression rTMS may resemble electroconvulsive therapy. As patients with depression have reduced activity in the left prefrontal cortex, high-frequency excitatory (brain stimulating) rTMS was tested. RTMS increased blood flow in this area (as measured with functional imaging), but it was not clear if this translated into functional improvement [12]. The idea that depression could also be caused by an imbalance of frontal lobe function, low-frequency inhibitory rTMS was tried on the opposite right prefrontal cortex. Effects were greater in patients without a history of psychosis, those responding to treatment with antidepressants and in younger people with younger age. However, as noted above, the investigators did not report changes of clinical relevance that were outlasting the stimulation. Hence, there is insufficient evidence to support the notion that rTMS is effective in the treatment of depression [12-14].

Administering an rTMS treatment to stroke patients may be considerably more promising. In this instance, there exists „clear evidence from experimental studies in primates as well as imaging studies in humans for reorganization“ [11]. It was shown that rTMS has an effect on reorganization in the natural restoration of cell functions which, though, crucially depends on the extent of the stroke. On the other hand, Khedr et al. state that rTMS administered in the initial two to four weeks after the stroke incident leads to a quicker recovery than in the case of patients treated with sham rTMS. An explanation for this finding could be that “changes in synaptic strength are the first stage in functional recovery” [11-12].

Common to many of these studies is the lack of sufficient knowledge of both the effective dose and the most suitable group of patients to treat. Ridding and Rothwell deduce that from these studies it is difficult to draw firm conclusions or gain more insight into how rTMS could be used for different

types of diseases. When considering the investigative effort spent on rTMS in its entirety, one might argue that 'in effect, the science has stood still' with respect to many diseases [11-12].

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3 Erklärung des Eigenanteils an der Publikation

Die vorliegende Doktorarbeit wurde in dem Journal „Brain Stimulation“ veröffentlicht und ist in dem Internetportal „Pubmed“ gelistet.
Der von mir zu dieser Publikationspromotion beigetragene Eigenanteil wird im Folgenden dargelegt.

Idee und Konzept der Studie:
Wurde von meinem Doktorvater entworfen.

Datenerhebung:
Nach Erstellung des Entwurfs, welcher durch meinen Doktorvater gemacht wurde, stand primär bei der Bewerkstelligung der Doktorarbeit die Datenerhebung im Vordergrund, welche ich nach Einweisung an den Geräten komplett selbständig durchgeführt habe.

Datenauswertung und Statistik:
Bei der Erstellung und Auswertung der Daten konnte ich durch die Expertise meines Doktorvaters das Erstellen und Auswerten von statistisch sinnvollen Tabellen erlernen. Dabei habe ich das Microsoft Programm Excel zur genauen Übersichtsdarstellung verwendet. Hier hat Herr C. Schwenke beratend gewirkt. Zur graphischen Darstellung der ausgewerteten Daten habe ich ein speziell dafür vorgesehenes Programm erlernt und folglich angewendet.

Interpretation und kritische Beurteilung der Daten:
Die Interpretation der Daten erfolgte einerseits durch meine Recherche in medizinisch-wissenschaftlicher Literatur und durch konstruktive Diskussionen mit meinem Doktorvater.

Manuskript:
Ich habe in Teilen den Manuskriptentwurf verfasst. Die endgültige kritische Durchsicht der Publikationspromotion wurde von meinem Doktorvater durchgeführt.

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5 Tabellarischer Lebenslauf

ZU MEINER PERSON

Persönliche Angaben

Maximilian Otto Fischer
Troppauerstraße 11
89257 Illertissen
Geboren: 09. Januar 1982 in Illertissen, Bayern
ledig, keine Kinder

CURRICULUM VITAE

Dissertation

12 / 2007 – 12 / 2013	Medizinische Dissertation an der Klinik und Poliklinik für Neurologie am UKE, Hamburg *Titel: Short-latency sensory afferent inhibition: conditioning stimulus intensity, recording site and effects of 1Hz repetitive TMS* *Leitung: Priv.-Doz. Dr. Michael Orth PhD*

Berufliche Ausbildung

Seit 09 / 2013	ATOS-Klinik Heidelberg, Orthopädie
11 / 2011 – 05 / 2013	Universitätsklinikum Heidelberg Department Orthopädie, Unfallchirurgie und Paraplegiologie – Paraplegiologie
05 / 2011 – 10 / 2011	Sana-Kliniken Lübeck Innere Medizin – Kardiologie

Akademische Ausbildung

12 / 2010	Prüfung über den zweiten Abschnitt der ärztlichen Prüfung (Staatsexamen)
10 / 2006 – 12 / 2010	Studium der Humanmedizin an der Universitätsklinik Eppendorf, Hamburg
06 / 2006	Prüfung über den ersten Abschnitt der ärztlichen Prüfung (Physikum)
09 / 2004 – 06 / 2006	Studium der Humanmedizin an der Universität Szeged, Ungarn
10 / 2003 – 02 / 2004	Studium der Rechtsmedizin (Jura) an der Universität Augsburg, Bayern

Praktisches Jahr (PJ)

08 / 2009 – 07 / 2010	*1.Tertial:* Allgemein- und Unfallchirurgie und Orthopädie im Hospital Argerich, Buenos Aires, Argentinien *2. Tertial, 1. Abschnitt:* Innere Medizin im Hospital Argerich, Buenos Aires, Argentinien *2. Tertial, 2. Abschnitt:* Innere Medizin am Unispital Basel, Schweiz *3.Tertial:* Neurologie am UKE, Hamburg

Famulaturen und Praktika

07 – 08 / 2008	Famulatur in der HNO und Radiologie am UKE, Hamburg
09 / 2007	Famulatur im staatlichen Lehrkrankenhaus Banjul, Gambia, Afrika
12 / 2006	Famulatur in der Allgemeinarztpraxis Dr. med. Günther Fischer, Illertissen
11 / 2006	Famulatur in einer Unfall- und Allgemeinchirurgischen Praxis in Augsburg, Bayern
03 – 06 / 2004	Krankenhauspflegepraktikum im Krankenhaus von Illertissen in verschiedenen Einrichtungen (OP, Innere Medizin, Ambulanz)

Heidelberg, 31.12.2013

6 Eidesstattliche Versicherung

Ich versichere ausdrücklich, dass ich die Arbeit selbständig und ohne fremde Hilfe verfasst, andere als die von mir angegebenen Quellen und Hilfsmittel nicht benutzt und die aus den benutzten Werken wörtlich oder inhaltlich entnommenen Stellen einzeln nach Ausgabe (Auflage und Jahr des Erscheinens), Band und Seite des benutzten Werkes kenntlich gemacht habe.
Ferner versichere ich, dass ich die Dissertation bisher nicht einem Fachvertreter an einer anderen Hochschule zur Überprüfung vorgelegt oder mich anderweitig um Zulassung zur Promotion beworben habe.
Ich erkläre mich einverstanden, dass meine Dissertation vom Dekanat der Medizinischen Fakultät mit einer gängigen Software zur Erkennung von Plagiaten überprüft werden kann.

Heidelberg, den 31.12.2013